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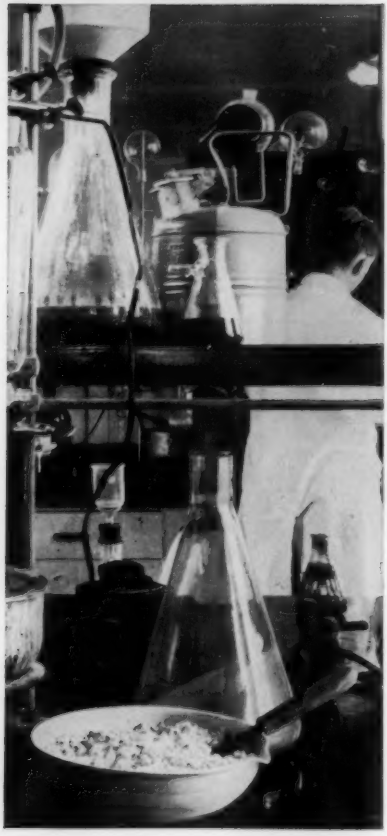

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OUR COVER

DR. JUSTIN L. POWERS

OUR cover this month is devoted to the best known and most highly respected editor and journalist in American pharmacy. Dr. Justin L. Powers had a distinguished career before joining the staff of the American Pharmaceutical Association but his many years as Editor of the Scientific Edition of its Journal and as Chairman of the National Formulary Committee have brought him into very close and intimate contact with the pharmaceutical scientists of this country and many others.

It is rare for a man to have both the ability to do the highly technical work which Dr. Powers has done and, yet, remain a close and good friend of all with whom he worked. This requires a skill and diplomacy not often found among those who have the scientific competence to do the kind of work which Dr. Powers has done, and done so well.

Dr. Powers in retiring from his official duties with the A.Ph.A. can reflect on a life of dedicated service to the profession which he loved so much. In his efforts to advance the cause and recognition of the pharmaceutical sciences, he also has the dual satisfaction of knowing that he has done this and, yet, leaves his official duties without having made a single enemy nor even a critic.

According to our standards, this is the epitome of successful living—to render effective service in one's work and to do it in a way which makes all those with whom you work your friends. The ovation given Dr. Powers by the Scientific Section of the Association and by the members of the Association itself at the last convention was an occasion of considerable emotion, deeply and sincerely felt not only by Dr. Powers but by his many long-time friends and associates.

We wish him well in whatever new responsibilities he may decide to accept. He will always be in our hearts and minds and among those whom we are proud to call our friends.

EDITORIAL

DRUG COUNTERFEITING

THE raid on the pharmaceutical company in Hoboken, New Jersey exposing all the sordid details of counterfeit drug marketing brought to light a shameful situation and one which should cause pharmacists everywhere great concern. Not only were *bona fide* prescription drugs being counterfeited and distributed as substitutes for legitimate prescription products but the conditions under which these drugs were made were almost unbelievable. Dirt, filth, and animal excreta were intermixed with chemicals and machinery and the published photographs are revolting to every pharmacist with any sense of self-respect or professional integrity. Those officials, both state and federal, who took action against this firm deserve credit for the forthright manner with which they acted in the public interest. The faulty and unsavory conditions within the plant were, of course, the most obvious area of criticism but other aspects of the case are even more significant and damaging to the prestige of pharmacy.

This is not the only company engaged in the counterfeiting of legitimate prescription drugs, as we feel certain any detailed and careful study would show. It has long been an open secret in pharmaceutical circles that such counterfeiting was going on and, indeed, there has been an active campaign on the part of legitimate manufacturers to reduce or eliminate this illicit traffic since, quite obviously, it is damaging to their economic interests as well as to public health. Here, it should be said that through cooperative effort on the part of boards of pharmacy, legitimate manufacturers, and the Food and Drug Administration considerable progress in this direction has been made in the last few years. The evil, however, has not been completely eliminated and, like all schemes based on human greed and avarice, it is not likely to be.

The most serious aspect of this problem has scarcely been touched upon by those who have discussed it in pharmaceutical journals and

before various pharmaceutical groups. Without question, that which should cause all of us the greatest concern is the knowledge that drug counterfeiting would not and could not continue another day if all pharmacists practiced their profession following strictly ethical principles and keeping public health and welfare uppermost in their minds. To condemn drug counterfeiting without condemning those pharmacists who patronize counterfeiters is exactly the same as condemning or imprisoning the lowly prostitute while paying no attention to those who patronized her. Pharmacists who buy counterfeit drugs know they are doing so every time. They also know that these counterfeit drugs are illegal, that they are of very dubious quality, and that no pharmacist would himself use such drugs or see them used by his family. The oft-repeated statement that pharmacists do this to save their customers money is a "dirty lie" since studies have shown that the customer for whom they profess such consideration and concern is usually charged the same as he would have been had he received the proper drug.

Practicing pharmacists cannot much longer endure these malpractices by their fellows who besmirch the name of pharmacy and push it still lower in public esteem. Those who love pharmacy and care for its future must insist on a system of self-policing which would see to it that any pharmacist guilty of such willful, unethical, and illegal practice be disbarred forever from his profession. For far too long now, pharmaceutical organizations have been too much in the nature of protective associations pledged to help the most scurrilous member out of difficulty even though it be deserved. Even some of our boards of pharmacy whose members in their oath of office pledge themselves to protect the public health and welfare seem loath to do so in many clear-cut cases, not based upon some technicality such as brand substitution but outright counterfeiting.

Pharmacy is presently engaged in a fight for survival and threatened on every side with controls and other changes which could spell disaster. We must establish a professional integrity which is beyond question and insist that all of our practitioners who wish to remain within the profession follow its ethical code of behavior to the letter.

L. F. Tice

EXPONENTIAL DE-EMULSIFICATION AND STABILITY OF PHARMACEUTICAL EMULSIONS

By E. Menczel, M. Rabinovitz and A. Madjor * †

A Reliable Colorimetric Method Is Described for the Determination of the Rate of De-emulsification Found to Be Exponential. The Stability of Pharmaceutical Emulsions Recorded Exponentially Enables Comparative Evaluation of Different Emulsion Systems.

THE stability of pharmaceutical emulsions is best ascertained by storage test methods measuring the time an emulsion persists (before it is split) at normal and accelerated conditions (1, 2). Husa and Becker (3) and later with Siragusa (4) combined the storage tests for pharmaceutical emulsions with the determination of dispersion decline of the internal phase through size frequency analysis (2, 5). It should be noted that this method could not be applied reliably to emulsions which are more stable in the coarse state of dispersion than in the fine one (5, 13).

The exponential nature of de-emulsification was suggested by Lederer (6) who has proposed an equation where the volume of separated internal phase is a function of time and velocity coefficient differing from one emulsion system to another. Ayres (7) has shown the mathematical relationship between exponential centrifugal and gravity sedimentation of emulsions. Cheesman and King (8) evaluated stability measurements of emulsions in log. seconds. Menczel *et al.* (9-11) determined the stability of pharmaceutical emulsions at exponential time intervals.

The rate of de-emulsification was measured experimentally by Tober and Autian (1) in a simple graduate cylinder whereas Cheesman and King (8) used an andiometer tube. The selectively soluble dyes, soluble only in one of the phases, are routinely used for the

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† We gratefully acknowledge the assistance given by Mr. N. Konforti of Hadassah Hospital, Tel-Aviv, in carrying out this study.

examination of emulsion types. Coloring emulsions with an exact amount of such dyes was evolved into an accurate colorimetric method for the determination of the volumes of separated internal phase at different times and thereby ascertaining the rate of de-emulsification.

Experimental

Emulsions

Non-stabilized oil-in-water and water-in-oil emulsions were colored with dyes soluble in the internal phase but insoluble in the external phase. Owing to the difference in the specific gravity of the interval and external phases, the distinctively colored internal phase could have been removed from the bulk of the emulsion without centrifuging through a separatory funnel. The composition of each of the selected emulsions was as follows:

	Emulsion			*Emulsion*	
	I	II		III	IV
Benzyl benzoate	← 25 ml. →		Lime water	← 25 ml. →	
Sudan IV	← 0.00125 Gm. →		Methylene blue	← 0.00125 Gm. →	
Sod. lauryl sulfate	0.3 Gm.	0.2 Gm.	Liquid petrolatum	72 ml.	65 ml.
Purified water to make	← 100 ml. →		Olive oil	3 ml.	10 ml.
Emulsion type	← o/w →		Emulsion type	← w/o →	
Specific gravity at 20°C.			Specific gravity at 20°C.		
Internal phase	← 1.117 →		Internal phase	1.018	1.018
External phase	← 1.001 →		External phase	0.890	0.900
Difference:	← 0.116 →		Difference:	0.128	0.118

A Westphal balance and a pycnometer at 20°C. were used for the determination of specific gravities. The type of the selected emulsions was verified microscopically.

The emulsions were prepared by the conventional method. Several 100 ml. portions of each of the prepared emulsions were distributed into conical separatory funnels (capacity 250 ml.) and re-agitated into a homogenous emulsion system. This has been considered as the 0 time of emulsion preparation. As soon as the separated internal phase of each of the emulsions could have been distinguished, it was collected from the separatory funnel and the time recorded. Later, further portions of the separated internal

phases were collected from the other separatory funnels containing the same but undisturbed emulsions indicating the time at each removal of the separated internal phases. Owing to the unequal emulsion splitting rate of these emulsions, this stepwise collection of internal phases was carried out at different times but, in each case, it was continued until practically the maximum separation was effected. The volume of the separated internal phase was determined according to its dye concentration by an evolved colorimetric method.

Colorimetric Method

Each of the separated volumes of internal phase was diluted up to 25 ml. with benzyl benzoate for emulsions I and II and lime water for emulsions III and IV. The concentrations of the dyes in such dilutions were determined colorimetrically according to the linear pre-calibration between the dye's concentration and optical density (Fig. 1). The colorimetric determinations were carried out in a Klett-Summerson photoelectric colorimeter which sensitivity was

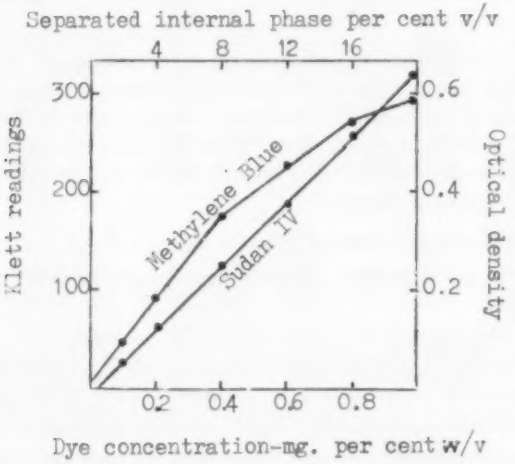

Fig.1. - Relationship between optical density (Klett readings) and dye concentration to percent of separated internal phase.

increased by a Kipp galvanometer (12) using blue filter-420 for sudan IV and red filter-640 for methylene blue. The readings were restricted to the sensitive range of the pre-calibration. It should be noted that the concentration of sudan IV in benzyl benzoate solution could be determined accurately by this method within the range of 0.1 mg. up to 1 mg. per cent, whereas that of methylene blue in lime water is within 0.1 to 0.4 mg. per cent. In case the scale readings of the colorimeter exceeded the sensitive range, the determinations were repeated upon proper dilution and, accordingly, the results were calculated. The concentrations of the dyes are directly proportional to the volumes of the separated internal phases and their per cent separation; accordingly, the following equations were derived for computing the volume and per cent separation of the internal phase:

$$V = \frac{C \times 25 \times d}{5} = 5\,d\,C \qquad \text{Eq. (1)}$$

$$V \text{ per cent} = \frac{C \times 25 \times d}{5} \times \frac{100}{25} = 20\,d\,C \qquad \text{Eq. (2)}$$

Where V is the volume of separated internal phase in ml., C its dye concentration in mg. per cent, 25 ml. is the original volume of internal phase which dye concentration had been 5 mg. per cent (0.00125 Gm. per 25 ml. of internal phase), and d is the dilution (footnote 1, Table I) in case the dye concentration exceeds the linear pre-calibration of dye conc. vs. optical density.

Rate of Separation of Internal Phase

Each of the emulsions exhibited three stages of internal phase separation:

a) Initial slow separation—about 25 per cent separation at:
0-25 hours (Emulsion I) 0-25 minutes (Emulsion II) 0-2.5 hours (Emulsion III) 0-45 hours (Emulsion IV)

b) Intermediate comparatively rapid separation—about 25-80 per cent separation at:
25-55 hours (Emulsion I) 25-65 minutes (Emulsion II) 2.5-6 hours (Emulsion III) 45-80 hours (Emulsion IV)

c) Final decreased rate of separation. In no case was complete separation of internal phase achieved.

Graphical evaluation of the rate of internal phase separation has revealed that the relationship between the per cent separated to the time is exponential: plotting both values as logarithms along the two axes, a straight line is obtained (Fig. 2) which follows the log-log coordinate equation:

TABLE I

THE VOLUME-TIME RELATIONSHIP OF LEAST AND UTMOST SEPARATION OF INTERNAL PHASE (EXPRESSED IN NON-EXPONENTIAL AND EXPONENTIAL UNITS)

	Emulsion I		*Emulsion II*		*Emulsion III*		*Emulsion IV*	
	Least Separation	*Utmost Separation*	*Least Separation*	*Utmost Separation*	*Least Separation*	*Utmost Separation*	*Least Separation*	*Utmost Separation*
Separated Internal Phase								
Dilution [1]	1:1	1:6	1:1	1:6	1:1	1:12	1:1	1:11
Optical density [2]	0.032	0.490	0.278	0.490	0.070	0.329	0.086	0.339
Dye-mg.p.c.w/v	0.05	0.766	0.44	0.766	0.08	0.376	0.09	0.387
Volume—ml.[3]	0.25	23	2.1	23	0.40	22.6	0.50	21.30
P.C.[4]—sep.[5]	1	92	8.4	92	1.60	90.4	2	85.2
log.$_{10}$p.c.[4] sep.[5]	0	1.963	0.924	1.963	0.204	1.956	0.301	1.930
In p.c.[4] sep.[5]	0	4.516	2.125	4.516	0.469	4.499	0.692	4.439
Time of Separation								
Seconds	1,800	216,000	600	7,200	1,800	28,800	43,200	288,000
log. 10 seconds	3.255	5.334	2.778	3.857	3.255	4.459	4.635	5.459
In seconds	7.487	12.269	6.189	8.861	7.487	10.347	10.661	12.556

[1] Dilutions: 1:1 is the procedural dilution of the internal phase with benzyl benzoate (Emulsions I and II) and lime water (Emulsions III and IV) up to 25 ml.; in case the dye conc. is beyond the sensitivity range of the photoelectric colorimetric pre-calibration—it was further diluted; 1:2 means up to 50 ml., 1:3 up to 75 ml. etc.

[2] The optical density is proportional to the dyes' concentrations in accordance with the linear calibration of fig. 1.

[3] The volume and per cent separated internal phase were calculated according to equations (1) and (2).

[4] Per cent v/v.

[5] Separated.

$$x = k\,y^{a} \qquad \text{Eq. (3)}$$

$$\log x = \log k + a \log y \qquad \text{Eq. (4)}$$

where y is the per cent separating internal phase; x, time in seconds; k and a are constants which differ from one emulsion system to another. The straight line in Emulsion I bends toward the end owing possibly to some inhibition of the de-emulsification at its last phase without affecting the net result (see the combining dotted-pointed line—Fig. 2).

Discussion

Exponential Emulsion Depreciation

The activation energy concept of de-emulsification is expressed by $N = N_o\, e^{-E/RT}$ where N is the number of emulsion globules having a high velocity to overcome E the energy barrier; R, gas constant; N_o, the total number of emulsion globules, and T, absolute temperature (14). It follows that the ratio $N/N_o = e^{-E/RT}$ could be converted into the exponential depreciation ratio of an emulsion expressed by $N_t/N_o = e^{-Z}$ where Z conforms to E/RT and accordingly to the multiple of rate of depreciation and time (t) corresponding to that of N_t. The exponential time measurements based on $\log_{10}$ (Fig. 2) were therefore converted into natural logarithm (upper side of x axis) and as well the $\log_{10}$ per cent separated internal phase into ln (right side of Fig. 2). The conversion of the least and utmost separation of internal phase (the two extreme points of the emulsions studied) are illustrated in Table I.

Perceptible De-emulsification Axis (P. D. A.)

Whatever the rate of separation of the internal phase, the de-emulsification process is at first non-apparent and later apparent. We might assume that the rate of de-emulsification is equal at both the non-apparent and apparent stages [positive and negative Y axis (Fig. 2)]; for pharmaceutical emulsions consisting of considerable volumes of internal phase dispersions, the separation of one (1) per cent internal phase ($\ln = 0$) renders the de-emulsification perceptible. The suggested perceptible de-emulsification axis (P. D. A.) is actually the demarcation boundary between apparent and non-apparent de-emulsification. The less the rate of de-emulsification, the more it will take to reach the perceptible de-emulsification axis (P. D. A.). If the slope of internal phase separation is drawn on upwards and extended into the non-apparent stage, the interception points (k)

with the perceptible axis of de-emulsification indicates the degree of emulsion stability. If the same exponential time units are used for the x axis as well as for the P. D. A., the higher the value of k, the

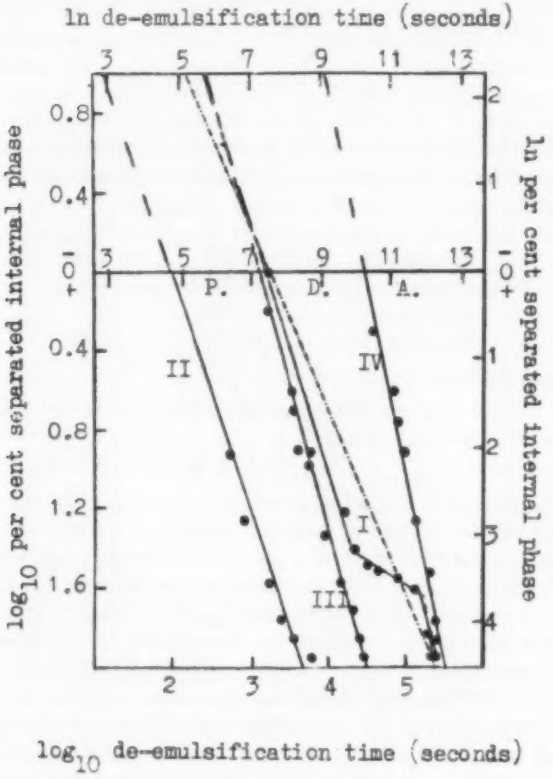

Fig. 2.- Rates of de-emulsification.
I - Emulsion I, II - Emulsion II,
III - Emulsion III, IV - Emulsion IV.
P.D.A. - Perceptible de-emulsification axis. ---- invisible rate of de-emulsification.

more stable is the emulsion. The value of k could be ascertained from Eq. (4) expressed in natural logarithm:

$$\ln x = \ln k + a \ln y \qquad \text{Eq. (4a)}$$

Solving this straight line question, k and a are computed by the conventional method:

$$x - x_1 = \frac{x_2 - x_1}{y_2 - y_1} (y - y_1)$$

The data tabulated in Table I could be used for these calculations and, accordingly, the emulsions investigated exhibited the following ln k and a:

	ln k	*a*
Emulsion I	7.487	1.06
Emulsion II	3.809	1.12
Emulsion III	7.154	0.71
Emulsion IV	10.215	0.50

Emulsion Stability Evaluation

It follows from eq. (4a) that when a $\ln y = O$ (at 1 per cent separation of internal phase)

$$\ln x = \ln k \qquad \text{Eq. (5)}$$

The value of ln k could be therefore considered as the exponential gradient of the decline in emulsion stability expressed in natural logarithm of the time at which 1 per cent of internal phase separated.

The tedious described procedure for the determination of ln k could be replaced by an alternative simple method. The freshly prepared emulsion is examined for the stability at exponential intervals, starting with base e (2.7183) seconds and onwards 2.7183^2, 2.7183^3, 2.7183^4 etc. at geometrical progression. As soon as emulsion instability is observed, the perceptible de-emulsification axis is actually reached and, if the time is converted into its natural logarithm, it will correspond to ln k of eq. (5). It is suggested that accordingly unique emulsion stability units could be established and defined as natural exponents of the apparent stability time of emulsion. For example: An emulsion stable for 22,000 seconds ($\ln 22{,}000 = 10$) but not at 59,870 seconds ($\ln 59{,}870 = 11$), the ln k value is somewhere between 10 and 11; however, this emulsion was definitely stable at ln 22,000 seconds ($= 10$) and, therefore, its stability could be

designated as 10 stability units (S. U.). The higher the numerical value of the stability units of an emulsion system, the greater is its stability (Fig. 3).

Since emulsion sedimentation is too exponential (7), it follows that, in case sedimentation whether positive or negative (15) precedes the separation of internal phase, it too could be recorded by the very same stability units adding the letters B, C, or D in parentheses. The letter B indicates the separation of two or more emulsified layers, C denotes significant sedimentation or its reverse (creaming), and D designates the separation of external phase from the bulk of emulsion.

The proposed exponential grading of emulsion stability supplements the hydrophile-lipophile balance (HLB) advocated by Griffin (16-19); thereby, homologous emulsion systems which acquired the proper and same HLB using different emulsifying agents could further be valuably screened employing these stability units (11).

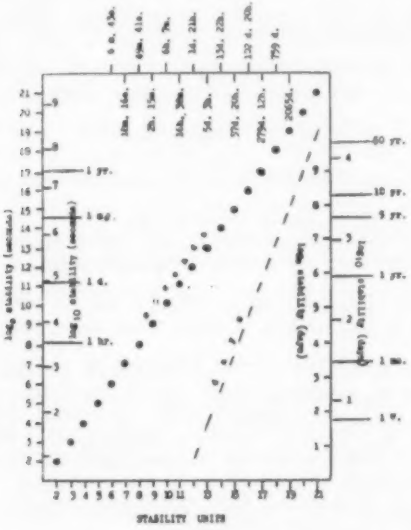

Fig. 3. - Stability units grading scale

s. - seconds, m. - minutes, h. (hr.) - hours,
d. - days, w. - weeks, mo. - months, yr. - years.

Summary

A photoelectric colorimetric method is described for the accurate determination of the rate of internal phase separation from o/w and w/o emulsions. The rate of internal phase separation was found to be exponential. Apparent de-emulsification has been differentiated from the non-apparent one by a perceptible de-emulsification axis corresponding to 1 per cent separation of internal phase.

Emulsion stability was evaluated by the intercept of the slope of de-emulsification with the perceptible de-emulsification axis. Accordingly, emulsion stability could be graded on the basis of exponential timely interval stability examinations. Simple numerical values designated as stability units (S. U.) express the exponential stability time to base e (in seconds) before de-emulsification is perceptible.

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SEQUESTRATION AND ITS APPLICATIONS

By John D. Connor *

IN the last twenty years, diversification in the use of sequestering agents has been expanded to such an extent that it may be truly said that any industrial user of water is a potential user of sequestrants. This rapid increase in utilization has been brought about mostly by a greater knowledge of their mode of action and an increase in the number of useful compounds now available. In this paper, we will attempt to give a brief outline of some of the information derived during this period.

Sequestration may be defined in the broad sense as the suppression of the reactivity of a metal in a system without actually removing the metal from the system or concentrating it in any part of the system. Such a definition rules out the inclusion of precipitants, extracting solvents, and ion exchange resins.

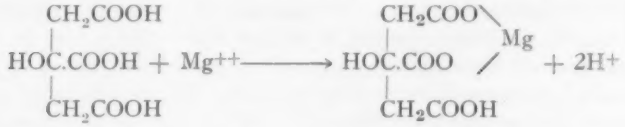
Although approximately ten general types of chemical structures have been found useful in sequestration, most of the consideration here will be given to the three groups with special technical significance at the present time. These are (in order of their introduction of use) the hydroxy carboxylic acids, the polyphosphates, and the amino carboxylic acids. All three of these groups function as sequestrating agents by means of chelation. A chelating (chele, Greek; a claw) agent is one which binds a metallic ion into a heterocyclic ring structure. Once bound in this non-ionic form, the metallic properties are completely altered. For example, a calcium chelate is not precipitated by ammonium oxalate. The metal will also be unable to catalyze some of those reactions which it normally would when in the free ionic state.

Hydroxy Carboxylic Acids

The most important members of this class have been citric, tartaric, and gluconic acids. Citric and tartaric acids, although not isolated until the latter part of the 18th. Century by the Swedish pharmacist, Scheele, occur widely in natural products and have been used in the crude form by chemists and, for that matter, housewives for centuries.

* Graduate student, Philadelphia College of Pharmacy and Science.

These hydroxy carboxylic acids form true chelate complexes, an example of which occurs in the preparation of magnesium citrate:



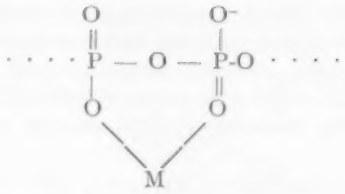
The magnesium ion, supplied in the poorly soluble carbonate form, is converted to a soluble chelate by incorporation into a five membered ring.

It has been suggested (1) that these acids have, in addition to chelate formation, a threshold effect which interferes with crystal growth by co-ordination at the crystal-liquid interface. This precipitation-retarding effect can be distinguished from true sequestration by a simple analysis. If the amount of calcium ions kept in solution by an agent in the presence of oxalate ions is the same as the amount of calcium oxalate which can be solubilized, the process is true sequestration. If the amount of calcium kept in solution exceeds the amount of calcium oxalate that can be solubilized by the agent, it is a threshold effect.

Hydroxy carboxylic acids have been used principally in the chelation of calcium, iron, copper, and lead. Their effectiveness varies widely depending on the metallic ion present, its concentration, and the pH of the system. It should be kept in mind that compounds such as these will support mold growth in aqueous solutions.

The Polyphosphates

The first sequestering compound of this class was discovered in 1833 by Thomas Graham, better known as the "father of colloid chemistry." This amorphous substance, later known as Graham's Salt, was prepared from sodium acid phosphate by removal of a molecule of water, followed by fusing and sudden chilling. Its exact structure is still unknown but the empirical formula given it is $NaPO_3$. It was once thought to be a salt of hexametaphosphoric acid, $(HPO_3)_6$. However, it probably forms long and complicated polymer chains. A divalent metal would be complexed thus:



The commercially important polyphosphates are pyrophosphate, tripolyphosphate, and the amorphous metaphosphates. These compounds, with various modifications, are sold under a variety of trade names. Their principal use is to soften "hard" water. A brief outline of their application for this purpose will be given here.

Hard waters are those containing objectionable amounts of dissolved salts of calcium and magnesium. These are present as bicarbonates, chlorides, or sulfates. These salts give insoluble precipitates with soap and form clogging scales with low thermal conductivities when used in boilers. Water hardness may be divided into two categories:

1. Carbonate or temporary, which can be greatly reduced by boiling.
2. Non-carbonate or permanent, which requires the addition of chemical agents.

Surface water supplied from watersheds over igneous rocks is comparatively soft. But, in recent years, it has become necessary to utilize the harder waters from limestone areas, especially in the Middle and Far West. The economic significance of plumbing occlusion and reduction of heat exchange is apparent. Each industry has its own water conditioning requirements; for example, laundries, textile manufacturers, and dyers require zero hardness to prevent precipitation of insoluble calcium and magnesium soaps (*2*). These divalent soaps not only increase the amount of monovalent soaps necessary, but also make the material stiff and subsequent dyeing spotty and off-shade.

The most important method for softening water is the cation exchange method, also known as the zeolite system, which employs natural or synthetic resin cation-exchangers. The lime-soda process is also widely used. Neither of these depend on true sequestration for their effect. The polyphosphates soften water by forming soluble

complexes with Ca, Mg, Fe, and Al ions. As little as 5 ppm results in prevention of after-precipitation and also possesses the valuable property of dispersing previously formed insoluble soaps (3).

Hall, the first producer of sodium hexametaphosphate for boiler water conditioning, theorized, in 1932, that his product ionized in two steps (4):

1. $(NaPO_3)_n \leftrightarrows 2Na^+ + Na_{n-2}(PO_3)_n^-$

and

2. $Na_{n-2}(PO_3)_n^- \leftrightarrows 2Na^+ + 2Na_{n-4}(PO_3)_n^{-4}$

The subscript $_n$ represents the number of $NaPO_3$ units in the polymer. Calcium ion may then be taken up in two ways:

1. $Ca^{++} + (NaPO_3)_n \leftrightarrows 4\ Na^+ + Na_{n-4}(PO_3)_n^{-2}Ca$
2. $2Ca^{++} + (NaPO_3)_n \leftrightarrows 6Na^+ + Na_{n-6}(PO_3)_n^{-2}Ca_2$

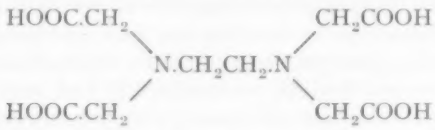
The number of Ca ions sequestered by one metaphosphate polymer is dependent on the value of n and the ionization constants of the complex negative ions formed.

Aqueous solutions of polyphosphates hydrolyze and deteriorate on standing, so their concentration must be carefully controlled by addition as needed.

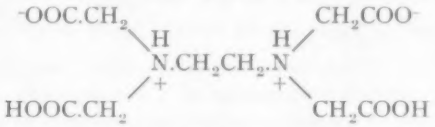
The Amino Carboxylic Acids

The outstanding current success of sequestering agents is due primarily to the versatility of the amino carboxylic acids and their salts. In 1862, Heintz produced amino triacetic acid from ammonia and monochloracetic acid (5); however, it remained little more than a laboratory curiosity for decades. Dr. F. Muntz, a chemist for the German dyestuffs combine of I. G. Farbenindustrie A. G., in 1935, was experimenting with various amino carboxylic acids and their effects on textiles. He found that amino triacetate was effective in preventing precipitation of insoluble salts and soaps of calcium and magnesium. In 1936, Farbenindustrie patented this product along with several related compounds, one of which was ethylenediamine tetracetic acid which was later to eclipse its companions (6). About 1950, coinciding with the post World War II industrial rebirth in Central Europe, the development of the potentialities of the amino-carboxylic acids began with Switzerland and Germany supplying the analytical theory, but with the United States as the point of greatest attack on its possibilities (7).

Ethylene diamine tetracetic acid or EDTA, which can be considered the classical example of this class, may be written as:



or as a zwitter ion (since it is a polybase as well as a polyacid):



It can be obtained in good yield by reacting ethylene diamine with chloracetic acid, or by the ethylene diamine-HCN-HClO-NaOH process. Another method is by catalytic oxidation of tetra (hydroxymethyl) ethylenediamine (8). It is a slightly soluble crystalline material which can be neutralized by up to four alkali metal cations to form a series of water soluble salts. It does not form anhydrides despite its polycarboxylic character. Strong heat causes molecular breakdown by decarboxylation. Its properties lie almost exclusively in the field of physical and co-ordination chemistry, as will be pointed out shortly.

Since EDTA is a tetrabasic acid, it can be thought to ionize in four steps, with four ionization constants. Considering the EDTA molecule as H_4Y:

$$H_4Y \leftrightarrows H_3Y^- + H^+ \qquad K_1 = \frac{(H^+)(H_3Y^-)}{(H_4Y)}$$

$$H_3Y^- \leftrightarrows H_2Y^{-2} + H^+ \qquad K_2 = \frac{(H^+)(H_2Y^{-2})}{(H_3Y^-)}$$

$$H_2Y^{-2} \leftrightarrows HY^{-3} + H^+ \qquad K_3 = \frac{(H^+)(HY^{-3})}{(H_2Y^{-2})}$$

$$HY^{-3} \leftrightarrows Y^{-4} + H^+ \qquad K_4 = \frac{(H^+)(Y^{-4})}{(HY^{-3})}$$

When EDTA is titrated with NaOH, the first two dissociation constants correspond to strong acids, and, if the titration curve is plotted, a sharp break is obtained after the addition of two alkali equivalents. Another equivalent may be taken up but, since K_4 corresponds to a very weak acid, it gives no sharp break in a titration curve. However, in the presence of excess calcium ions, all four protons are dissociated readily, the calcium coordinating with both nitrogens, forming CaY^{-2}.

It is important to remember that the reaction

$$\text{Chelate} + \text{Metal Ion} \leftrightarrows \text{Metal Chelate} + \text{acidity}$$

is reversible (9). If acid is present in excess, the reaction comes to equilibrium by increased production of the compounds on the left. On the other hand, if the acid is neutralized as quickly as it is formed, the equation products on the right will predominate. It is not difficult to deduce then that chelating agents are more efficient in an alkaline medium. Below a pH of 7.5, calcium and magnesium are not complexed efficiently. Copper, however, can be chelated almost completely at pH 3.5. Chelation will then be preferential under varying degrees of acidity. A solution containing a mixture of metals at a certain pH may have one metal sequestered leaving the other unchanged. Use of this principle is made in the isolation of rare earth metals and in selective removal of offending ions in analytical chemistry. Paradoxically, a high pH can also introduce difficulties since hydroxyl groups are precipitating agents for many metals, particularly ferric iron. It is generally conceded that it is more feasible to prevent a precipitate than to redissolve one.

The increase in acidity noted above can be used to determine the concentration of a chelate in solution by ordinary electrometric titration. In general, the increase in acidity on complex formation follows the increase in stability of the complexes formed.

The extent to which complex formation occurs may be expressed by an equilibrium constant K for the reaction:

$$\text{Metal}^{++} + \text{EDTA} \leftrightarrows \text{Metal-EDTA}$$

$$\text{where } K = \frac{(\text{MeEDTA})}{(\text{Me}^{++})(\text{EDTA})}.$$

K then is the ratio of metal which occurs as (MeEDTA) to the metal present as the ion. Where K is large, the metallic ion concentration will be low. EDTA forms complexes with all the alkaline earths,

heavy metals, and rare earths and even weak complexes with Na^+ and Li^+. The reciprocal of the equilibrium constant (1/K) is the dissociation constant for the complex.

Metal ions vary in the ease with which they combine with EDTA, as indicated by the K of various metal complexes expressed logarithmically in a 0.1N KCl solution at 20°C. (10):

	log K		*log K*
Na	1.66	Zn	16.58
Mg	8.69	Pb	18.2
Ca	10.59	Cu	18.38
Fe(ous)	14.22	Ni	18.45
		Fe(ic)	25.

The greater the log K value, the more stable the complex and the more efficient will be chelate formation.

EDTA and its di-, tri-, and tetrasodium salts are available under names such as Sequestrene (Geigy), Hampene (Hampshire Chem. Corp.), Perma-Kleer (Refined Prod. Corp.), and Versene (Dow). As the name of this class (amino carboxylic acids) implies, there are numberable organic compounds besides EDTA and its salts which are used and can be placed in this category (11).

Other Types of Sequestering Agents

In this section will be introduced some other chemical structures which have been found useful but which have not as yet achieved the stature attributed to those discussed previously.

Schiff's Bases is the name designating a group of compounds typified by disalicylaldehyde ethylene diamine. They are relatively unstable compounds which are made more stable through complexing. One particular advantage of this group is their oil solubility. This makes them especially useful in the petroleum industry.

Amino alcohols or ethanolamines have been known to possess chelating properties for some time. However, sequestration occurs only under strongly alkaline conditions.

Unidentate (one point of attachment) ligands such as fluoride or acetate ions rank as the least expensive of the sequestering agents. They are often employed in analytical processes.

Often employed in extraction solvents, 1-3 diketones are potentially within the sequestrant class.

Practical Application

The uses of sequestering agents, no matter what the field of endeavor, may be classified under four main headings:

1. Prevention or redissolving of precipitates
2. Suppression of the ionic form as an interfering entity
3. The use of a metal in its chelated form
4. The stabilization of a particular metallic valence state.

As a precipitate preventive, the sequestering agents are used in water softening, photographic developers, textile and dyeing processes, and as clarifying agents in liquids such as shampoos, liquid soaps, wines, vinegar, hair waving lotions, weed killers, and so on. It is interesting to note that the cloudiness seen in some bottled and canned liquids on standing is due to the leaching of metallic ions from the container. For example, liquid shampoos are capable of extracting calcium from glass. In some cases, this just results in an unsightly product; in other cases, the active ingredient may be partially or wholly degraded.

The suppression of ions which interfere in production can take many forms. Trace metals cause deterioration of ascorbic acid and increase the rancidity rate of foods containing fats and oils. Bleaching agent additives in detergents are stabilized against breakdown until their ultimate washing use by the elimination of trace metal catalysts.

Another of the practical applications is the use of the metal in the chelated form. It is quite possible that the chelate serves as a reservoir of a low but renewable concentration of simple metallic ion. In this connection, cyanide ion complexes are most efficient in providing CN^- for industrial electroplating. In agriculture, metals needed for proper plant metabolism may be unobtainable, either due to their deficiency in the soil or their being present in an unassimilable form. If a metal such as copper is applied in elemental or salt form in large concentrations, it may injure or kill the plant. However, chelated metals, when sprayed on the foliage or injected at root level, have been most successful in preventing or reversing chlorosis.

Calcium di-sodium EDTA has been employed medically in heavy metal poisonings, the mechanism to be discussed later under physiological applications.

The last application, that of complexing and sustaining the various states of metals, has become increasingly important in recent organic syntheses. Some cations are too unstable to exist unless there is appreciable co-ordination support. Not only does EDTA supply co-ordination bonds, but it also has the tendency to stabilize a higher valence state at the expense of a lower. In the table given previously, it can be noted that the log K value of ferrous iron EDTA chelate is 14.22 while that of ferric iron is 25. As mentioned before, the greater the log K value, the more stable is the complex. Ferrous EDTA chelates are sufficiently powerful as reducing agents to reduce iodine to iodide ions. This makes them useful in commercial photography as reducing agents. The action of ferrous iron on peroxide-initiated vinyl polymerization is assisted if the iron is in an EDTA chelate form (12). This has been used in a number of "Redox" recipes for emulsion polymerization.

Throughout this article, one or more uses for sequestering agents in the following industries have been discussed: agriculture; chemical production; cosmetic; food and beverage; heat transfer equipment; water conditioning; metal finishing; mineral separations; paper; photography; rubber, polymers and plastics; soaps and detergents; textiles; fats and oils; analytical chemistry. A separate category has been reserved for discussion of their use in relation to health.

Physiological and Pharmaceutical Applications

In considering this section, it may be well to recall that sequestration complexes are not foreign to biological systems. Chlorophyll, cytochrome, and blood pigment are typical examples found in the plant and animal kingdoms.

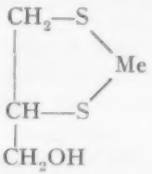
Perhaps because of a failure to understand the basic underlying principles involved, the progress made in the utilization of sequestering agents in medicine and associated fields has been by trial and error in many cases. Nevertheless, some important uses for these agents have evolved and more will certainly follow.

Toxicity studies conducted with the three important classes of sequestering agents show a low order of toxic manifestations. Citric and tartaric acids are present in the average diet while the polyphos-

phates are readily split *in vivo* by enzymes. Since EDTA forms a stable compound which is foreign to the body, it has been studied extensively for toxic possibilities. Patch tests and intradermal injections have demonstrated that it is no more irritating than an equivalent amount of soap solution. Oral toxicity was checked on a group of rats given EDTA in doses equivalent to two to five per cent of total diet intake (food and water). At the end of a year, there were no untoward manifestations (13). In the same experiment, a concentration of 100 mg./kg. by rapid intravenous injection was required to produce hypocalcemic tetany in rabbits. Two Gm./kg. were required to produce death when given by intravenous infusion over a three hour period.

Information based on these and similarly conducted tests indicate a high level of safety under most conditions and warrant the use of these agents in materials to be ingested by or injected into living organisms. The Food and Drug Administration has, from time to time, given its permission for the inclusion of sequestering agents in products under its jurisdiction.

In cases of lead poisoning, EDTA has been found more effective than 2,3-dimercaptopropanol (BAL). It is given by slow injection in the form of the calcium disodium chelate. Since the log K value of the lead chelate is greater and therefore more stable than that of the calcium chelate, the calcium is released and the lead taken up. This soluble lead complex is then excreted by the kidneys. BAL, which can also be considered a chelating agent since it forms a soluble five-membered ring with heavy metals,



remains the drug of choice in mercury poisoning. The use of calcium chelate in lead poisoning prevents production of concurrent hypocalcemia. However, the injection of the disodium form of EDTA has been tried in hypercalcemic states with varying reports as to its utility for this purpose.

Unwanted metallic deposits in the body such as calcified corneal deposits and urinary calculi have been treated by direct application of EDTA or retrograde irrigation. In many cases, this course of treatment was abandoned due to ensuing irritation (14).

Oral administration of ferric sodium EDTA for the treatment of hypochromic enemia has been described (15). Serum iron levels compared favorably with those achieved with ferrous gluconate without the gastrointestinal side effects frequently associated with the latter. Aqueous preparations of the trivalent iron chelate are stable which permits the marketing of palatable liquid dosage forms. The trivalent iron is believed to be reduced to a soluble divalent form in the stomach and duodenum.

Mention has been made that EDTA or its calcium chelate, when administered orally or by injection, will limit cholesterol plaque formation by lowering the serum cholesterol and low density beta lipoprotein levels. Current reports (16) refute this.

Attempts have been made to add EDTA to dentifrices in order to increase calculus removal and to inhibit *Lactobacillus acidophilus* which may be one of the causes of dental caries. Unfortunately, even the calcium chelate form does not prevent the agent from stripping calcium from the teeth.

Heavy metal chelates which are radio-opaque can be concentrated in certain organs for X-ray purposes with less toxicity than other forms of the metal (17). One such compound has been patented.

The use of EDTA as an anticoagulant for blood has been recommended. Blood so stabilized can be used for whole blood transfusions or plasma extraction, whichever is later desired.

One or more of the three classes of sequestering agents have been found to stabilize and retard decomposition of solutions of tetracycline, crystalline penicillin, and Vitamins B_{12} and C. Trace metal-induced oxidation and discoloration are prevented in solutions of sodium salicylate, sodium-p-amino salicylate [which is also made less irritating to the gastric mucosa without impairing its tuberculostatic activity (18)], antihistamines, sulfadiazine, epinephrine, rutin, and the barbiturates.

Hard water has an adverse effect on quaternary ammonium bactericides. This is not due to any precipitating or deactivating mechanism. Most bacteria carry an anionic charge, and metal ions present in solution will compete with the quaternaries for deposition

on the bacteria. The polyphosphates or amino-carboxylic acids should be present to prevent this interference.

Much interest has lately been directed toward the use of sequestering agents in physiologic and industrial enzymatic processes. Enzymes, being of a proteinaceous character, undergo many involved molecular changes such as methylation, carboxylation, and deamination while performing their specific catalytic function. Some of these group rearrangements require the presence of metallic ions. Sequestering agents may then act (19):

1. As inhibitors by removal of the metal from a prosthetic group or
2. As activators or stabilizers by removal of metal ions blocking essential groups and acting as non-specific inhibitors.

Recent studies in the field of radiation decontamination have shed some light on a new and exciting area of possible utilization. As might be expected, sequestering agents in combination with detergents will quickly remove strontium^{90} and other radioactive metals from the skin. Indeed, solutions of EDTA have been employed for some time to decontaminate personnel, animals, equipment, and buildings exposed to radiation. However, research at the Jewish Hospital in Brooklyn has shown that, unlike calcium, strontium will not form osseous nuclei from which bone and tooth crystals grow (20). It was further found that, although strontium concentrates in bone tissues, it concentrates on the outside of the already formed calcium phosphate nuclei. Certain chelates, notably calcium bis-[(dicarboxylmethyl) amino ethyl] ether, increased the urinary excretion of radiostrontium in rats given 0.6 to 0.8 microcuries of Sr^{85}. When chelates were given immediately after exposure there was a significant increase in excretion of radiostrontium, both in the ionic form and as SrBAETA chelate over a twenty-four hour period. Since the excretion noted falls off rapidly with an increase in the time between exposure and treatment, prophylaxis prior to exposure will be the major application. Apparently, selected stable strontium, calcium, or magnesium chelates participate in an ion exchange equilibrium with available radio-active cations in bone, this shift in equilibrium favoring the excretion of radiostrontium.

Conclusion

It should be evident at this point that the sequestrational art has advanced considerably in a short period of time. What was until recently a little-understood subject of theoretical musing has become responsible for important industrial evolutions. The production of these agents has become in itself an important facet of our economy. From all indications, the application of sequestering agents to solve old problems and open new fields is limited only by the abilities and imagination of man.

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A DECADE OF MANAGEMENT OF POLYCYTHEMIA THROUGH CHEMOTHERAPY

John R. Sampey *

RADIOPHOSPHORUS has long dominated the therapy of polycythemia. In this survey of more than 4,000 cases during the last decade, 3662 were treated with P^{32}. A statistical evaluation of the results of the treatment with this radioactive isotope is difficult, for a dozen clinical reports fail to state the number of cases treated, and 23 of the 98 published studies do not give the number responding.

Triethylene melamine, nitrogen mustards, and folic acid antagonists are commanding increasing attention in the control of polycythemia. Karnofsky (47) rates P^{32}, N-mustards, and TEM as the most effective agents in treating this malignant disease, while Ariens (2) ranks TEM below P^{32} and urethan. A dozen miscellaneous chemicals have been tested in the control of polycythemia, and some of these give promise of more extensive use. Myleran, phosphoramides, hormones, colchicines, and antibiotics have been employed widely in related neoplasms (84-86).

Radiophosphorus

An evaluation of the 98 clinical reports on P^{32} therapy of polycythemia is beyond the scope of this study. Attention is directed to the fact that, of the 3662 patients treated with this isotope (Table I), 1343 were not evaluated in the 23 published reports for the authors gave the number of patients undergoing treatment without stating the number responding. Frequent descriptions are made of the more striking responses, along with discussions of the complications arising during the treatment.

Much has been written on the risks of radiophosphorus therapy of polycythemia. Bjerkelund (10) recommended the use of P^{32} only when other methods failed, because of the danger of radiation damage. Reinhard and Hahneman (77) tabulated nine deaths due to acute leukemia and five hemorrhages in 98 patients, with five more developing chronic myeloid leukemia and seven, splenomegaly.

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Stroebel (93) described the death of several patients with acute leukemia, and serious hematologic reactions in others following radiophosphorus treatment. Chodos and Ross (13) recorded the death of one subacute granulocytic leukemia out of seven polycythemia patients treated with the isotope. Currently, Masouredis and Lawrence (104) have discussed critically the problem of incidence of leukemia in radiophosphorus therapy.

Triethylene Melamine

The effectiveness of triethylene melamine in the control of polycythemia has been pictured in two dozen clinical reports. Rosenthal and Rosenthal (80) secured 12 good remissions, eight fair to good, and six fair to poor responses in 30 patients; their longest remission was 14 months, but 10 patients were alive at the time of reporting. In another study (79), these investigators reported satisfactory clinical and hematologic responses in 20 of 30 cases. Best and Limarzi (105) treated 30 patients with TEM but described only one case, a 15 month remission; they rated TEM next to P^{32} in the management of polycythemia. Linke and Lasch (59) treated 15 patients with a combination of TEM, urethan, and N-mustard; they recorded that the blood picture was normalized in 14 cases with remissions lasting from five months to three years. In a second study (58), they reported that nine patients responded well to TEM. Ellison and associates (20) noted hematological improvement in six patients accompanied by complete clinical response in three and partial, in two cases. Wasserman and Rashkoff (100) described good palliation in some of 17 patients on TEM; only one failed to respond, but one had severe anemia develop on the therapy; in a review article, Wasserman (101) expressed a choice of P^{32} over TEM and N-mustard in the management of polycythemia.

Lawrence and associates (53) observed remissions for over a year in two of seven patients on TEM, and two more had responses of less than a year. TEM gave good results in three of four patients described by Haga *et al.* (33) and Hall and Rheingold (34) reported that three patients were doing well at the time of reporting after TEM treatment. Rundles and Barton (82) judged the action of this drug as inconclusive in three patients. Walsh *et al.* (98) classed one result as excellent and another as fair in two cases treated with TEM, and Hansen (37) reported that one of two patients

showed some improvement on the drug. Louis and collaborators (60) recorded that three of 23 patients had 2-4% decrease in hematocrit on TEM, eight had 5-10% decrease, one had 22% decrease, five showed no change, and one an increase of 6 per cent.

Good remissions have been reported in single cases of polycythemia on TEM therapy by Haex *et al.* (32), Hambly and Robertson (35), Melamed (66), Rundles (83), Winkler and Cerny (102), and Yanguas (103). Some palliative results have been recorded in single cases by Baserga *et al.* (5), Colsky *et al.* (15), Hamerman and Melamed (36), Pavlovsky and Vilasega (74), and Schulze *et al.* (88).

Nitrogen Mustards

Klima and Hornischer (51) reported five good remissions, six fair and one doubtful response in 14 patients with polycythemia who received the nitrogen mustard, HN2. Bauer and Erf (6) reported that 12 of 14 patients were living at the time of reporting, but they considered HN2 as a dangerous drug in treatment of polycythemia. Notice has already been taken of the report of Linke and Lasch (59) on the combination therapy with N-mustard, TEM, and urethan in the management of 15 cases of polycythemia. Spurr *et al.* (91) recorded an average period of remission of 6.5 months in 10 cases on HN2 therapy. Shullenberger and Watkins (89) described some remission in all eight patients on this mustard. In 1951, Iverson and Meulengracht (45) observed improvement in five of six patients given the aromatic mustard, R48 (β-naphthyl-di-2-chloroethylamine) and, the following year (46), they reported some remission in a clinical study of six other patients with this mustard. Barlow *et al.* (3) described remission in three of four patients lasting three, four, and six months on treatment with mannomustine (1,6-bis(β-chloroethylamino)-1,6-deoxy-D-mannitol dihydrochloride). Piper (75) induced complete hematological remissions in three patients with R48, one lasting 18 months. The other continued in remission after 24 months. Colarusso and Boni (14) observed fair remissions of 15 and 16 months in three patients after HN2 therapy, and Leibetseder and Rabl (54) obtained three good responses with this mustard. Videbaek in two studies (94, 95) reported good remissions in four cases of polycythemia with N-mustard, and Goldeck (29) noted some improvement in two cases. Good responses have been recorded in single cases by Bisaro and Butto (9), Ebbe (19), and Mathews (64)

by treatments with HN2 or R48, and fair responses in single cases by Del Dotto and Scardigli (17), Graulich (30), Mocchi (69), Schatter (87), Sperlich (90), Warter *et al.* (99), and Rider and Warwick (78). Brief responses were noted by Fleischhacker and Klug (25) and Holy and Suchan (41) after combination X-ray and HN2 or HN3 therapy. Good palliation was noted by Iverson and Meulengracht (44), Klima and Hornischer (52), and Videbaek (95) in patients administered N-mustard.

Folic Acid Antagonists

Bell and Davis (8) employed the folic acid antagonist, amino-anfol (4-aminopteroylaspartic acid) in one patient with polycythemia, and noted a favorable clinical response. Isaacs (43) reduced the red blood cell count and observed amelioration of the symptoms of polycythemia by treatment of six patients with Daraprim, an antimalarial drug with antifolic acid properties. Kleinfelder made two studies on the action of Daraprim: in 1956, he and Brochere (49) observed good palliative response and reduction of both erythrocytes and leukocytes in six patients and, three years later, he described good control to three years in nine cases, but he recounted two had hemorrhages (50). In 1958, Frost *et al.* (28) conducted clinical trials with the folic acid derivative, pyrimethamine, on 14 patients: they described a fall in hemoglobin and red cell count in 11 patients, with a reduction in the size of the spleen and relief of symptoms. Four years earlier, Frost and Jones (27) noted some remission in five patients after treatment with 2,4-diamino-5-p-chlorophenyl-6-ethyl pyrimidine. Marchal *et al.* (62) observed severe anemia in a patient receiving pyrimethamine and Palou *et al.* (71) noted that this drug reduced the red cells to normal in one patient.

Miscellaneous Agents

Hule (42) and Fenicia (24) both reported some palliation in three patients with polycythemia following treatment with urethan. Florijn (26) described a patient with combined chronic myelosis and polycythemia who experienced a good remission on urethan. Feher (23) induced some remission in patients with polycythemia and leukemia with urethan. Rosenthal (81) noted symptomatic improvement in one patient on urethan and the corticosteroid, triam-

cinolone. Attention has already been directed to the combination therapy of urethan, TEM, and N-mustard.

Lawrence and associates (53) reported one remission lasting over a year and two lasting less than a year in three patients on phenylhydrazine therapy. Falconer (21) described a patient who was maintained for 19 years on phenylhydrazine. Haez *et al.* (32) noted good response in two patients on phenylhydrazine alone and in one on a combination of this drug and P^{32}. Levrat *et al.* (57) found a combination of phenylhydrazine and X-rays gave a poor response in one patient with polycythemia. Plauchin *et al.* (76) reported acute anemia during phenylhydrazine treatment of one patient, and Keesal and Margolis (48) warned of kidney damage by the drug in another case.

Louis (61) reduced the hematocrit readings in 18 patients to normal for as long as 65 weeks with myleran (busulfan). Wald *et al.* (97) secured some remission (7 complete) in five patients with myleran; all had one or more remissions which lasted to 19 months. Hansen (37) observed some response in one of two patients on this drug, and Dameshek *et al.* (16) reported promising preliminary results in his clinical trials on polycythemia.

Greer (31) induced a seven months' remission of polycythemia which was complicated by alcoholic cirrhosis and chronic myeloid leukemia, by large doses of ascorbic acid. Merlini and Torcicliani (67) noted the destruction of red blood cells in a patient given this vitamin in combination with alloxan. Hinz (40) recorded that vitamin B_{12} was without effect on a patient with polycythemia following pernicious anemia.

Block and associates conducted two clinical trials on radioarsenic: in 1949 (11), they described one three months' remission and, in 1953 (12), they induced a fair response in a patient with As^{76}. Beickert and Jorke (7) noted five good remissions in polycythemia patients receiving thiocyanate. Haynal published two papers on the use of p-hydroxypropionone on patients with polycythemia: in 1953 (38), he observed the improvement in red blood cells and hemoglobin in four patients and, in 1955 (39), he obtained a good response in 10 of 24 patients. Leonard *et al.* (55) caused some remission in 14 cases who were placed on triethylene thiophosphoramide therapy. Levrat *et al.* (56) described a hematological remission in a patient with polycythemia coexistant with a slowly progressing cancer of the

prostate: they administered distilbene. Meythaler and Händel (68) induced four good remissions in five patients with a combination of X-rays and hormones (Pancortex, Phenosol, etc.). Paterson and Boland conducted two clinical trials on the action of the ethyleimine, M9500, on polycythemia: in 1951 (72), they described two good clinical remissions without hematological response and, in 1953 (73), they rated as only fair the action of the drug on one case. Storti and Gallinelli (92) found Substance F, an extract of colchicine autumnale, gave some relief in one patient. Barnard *et al.* (4) employed massive oral doses of streptomyces-derived B_{12} therapy in two patients with polycythemia; both returned to work and were being maintained at the time of reporting by use of terramycin therapy.

Summary

Table I summarizes the chemotherapeutic management of polycythemia. Column 3 tabulates the number of cases which are included in Column 2 but for which the authors did not state the number responding. In seeking to evaluate "good" and "fair" remissions, three factors were considered: (1) Degree of hematological response; (2) Clinical improvement, both objective and subjective; (3) Length of the response. The large proportion of good remissions in P^{32} and TEM therapy is gratifying, and the high remission rates indicated with N-mustards and folic acid antagonists should stimulate further clinical testing of these chemicals.

Acknowledgment

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TABLE I

MANAGEMENT OF 4090 CASES OF POLYCYTHEMIA

Chemicals	*No. of Cases*	*Cases Not Evaluated*	*Good Remissions*	*Fair Remissions*	*Rem. Rate*	*No. of Refs.*
P^{32}	3662	1343	1078	681	76%	98
TEM	192 *	29	71	57	79%	25
N-mustards	87	—	18	62	92%	29
FAA	42	—	9	30	93%	8
Miscellaneous	107	—	47	40	——	29

* 15 patients were on combination therapy with N-mustards, etc.

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BOOK REVIEW

The Story of Chemistry. Georg Lockemann. 277 pp. Philosophical Library, Inc., 15 East 40th St., New York, 1959. Price: $4.75.

This new history of chemistry consists of two devisions of the science—the first relating to the writings and theories from antiquity, through Alchemy, Iatrochemistry and the Phlogiston era.

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Much of the book is concerned with inorganic chemistry and the many theories and legends leading to the establishment of chemistry as a true science.

There is much information concentrated in this volume which would be difficult for one not versed in historical literature to obtain elsewhere. A bibliography of some sort would have made further investigation of the original literature available to those interested in particular phases of the history of chemistry.

An index of names of investigators, mentioned in the text, completes this concise, but interesting, view into the events which established chemistry as a distinct science and of the men who devoted their thinking to further its cause.

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